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Dr Juneja and colleagues reply as follows:

We are surprised that Dr Cazzola should have found the conclusions confusing, despite our attempt to be as objective as possible. As mentioned, the cases in our study were classified according to the FAB proposals.¹ Accordingly, any case of a myelodysplastic syndrome with $\geq 5\%$ blasts in the marrow would be classified as refractory anaemia with excess of blasts. It is also mentioned in the FAB proposals that ringed sideroblasts may be seen in refractory anaemia with excess of blasts and no limit on their percentage has been laid down unlike that for acquired idiopathic sideroblastic anaemia. Moreover, it has been shown that the percentage of marrow blasts and not the ringed sideroblasts is the more important factor that correlates with prognosis in the myelodysplastic syndrome.² It is, therefore, important that the cases of myelodysplastic syndrome with excess ($\geq 5\%$) marrow blasts irrespective of the percentage of ringed sideroblasts be classified separately and not included in the group acquired idiopathic sideroblastic anaemia, the cases of which according to the FAB proposals, have a normal ($<5\%$) marrow blast count. Likewise it may not be justified to classify a case of myelodysplastic syndrome with 20-30% marrow blasts and 30% ringed sideroblasts as acquired idiopathic sideroblastic anaemia. All these points are discussed in greater detail in our second article³ which will be published shortly.

We also wish to point out that Cazzola *et al.*⁴ defined the cases of refractory anaemia with excess of blasts on the basis of the

percentage of marrow blasts and promyelocytes (10-30%), whereas we defined these on the basis of the percentage of blasts only, in the marrow (5-20%) and in the peripheral blood ($<5\%$), as in the FAB proposals. Therefore, the diagnostic criteria in the two studies were not identical. Even if we take the criteria of the blasts and promyelocyte together for defining refractory anaemia with excess of blasts, as in the study by Cazzola *et al.*, we still find eight cases which have $>30\%$ ringed sideroblasts and 19 cases which have $>5\%$ ringed sideroblasts.

Like Cazzola *et al.*, we appreciate the difference between the intermediate (or ferritin) and the ringed sideroblasts and only cells meeting the criteria of five or more Prussian blue positive granules covering one-third or more of the circumference of the nucleus were included, in our study.

We agree that the recent FAB proposals are a step forward, but we feel that they should not be modified until there is sufficient evidence to the contrary.

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Evidence for polymorphism of transferrin receptors in human skin

It was with considerable interest that we read the paper on transferrin receptors in human tissues by Dr Gatter and colleagues in the May 1983 issue.¹ In light of the poss-

ible importance of these receptors we feel it is important to communicate that our findings in normal skin with the use of monoclonal anti-transferrin receptor sera and immunofluorescence methods are at variance with those of Gatter *et al.* They reported transferrin receptor-positive cells in the basal layer of the epidermis using four monoclonal antibodies to transferrin receptors studied with the aid of the immunoperoxidase technique.

Using cryostat sections of normal full-term human placenta as the control tissue for normal human skin, we have carried out experiments which fulfil the necessary criteria for the demonstration of transferrin receptors.² These are: (a) direct immunofluorescence with the use of tetramethyl rhodamine isothiocyanate conjugated goat antihuman transferrin;³ (b) direct immunofluorescence, as above, following preincubation with 50 μ l of 2.5 mg/ml transferrin in phosphate-buffered saline (PBS);⁴ and (c) indirect immunofluorescence by using the immunoglobulin (Ig) fraction of rabbit antihuman transferrin and fluorescein conjugated sheep antirabbit Ig.⁵ These reactions were consistently negative in normal skin.

The monoclonal antibody OKT-9 has been well characterised as recognising transferrin receptor.⁷ In none of our experiments did this antibody react with cryostat sections of skin when used with fluorescein isothiocyanate conjugated goat anti-mouse IgG. However linear positivity was found uniformly on the syncytiotrophoblastic plasma membranes of chorionic villi (Figure).

It is apparent that several monoclonal antibodies which recognise transferrin receptors differ in their patterns of reactivity. For example 83/25 reacts with T, B and non-T, non-B cell lines⁸ and OKT9 preferentially recognises acute lymphoblastic leukemia T cells.⁹ In addition it has been shown that L5-1 primarily reacts with erythroid precursor cells.¹⁰ It is possible that the transferrin receptor is a family of structurally related but antigenically and functionally distinct molecules such as was suspected as a result of studies of breast adenocarcinoma cells.⁵ This is borne out by the present observations which show that monoclonal OKT-9 antibody does not recognise basal cells in human skin, but the monoclonal antibodies to transferrin receptors used by Dr Gatter and colleagues gave a quite different result. Barring some type of unforeseen technical artefact, we interpret these results as being another manifesta-

The distribution of transferrin receptor on human placenta. The syncytiotrophoblast of chorionic villi is positive with monoclonal antibody OKT-9. Note negativity of chorionic stroma (S). $\times 300$.

tation of the polymorphism of human transferrin receptors. At present it would seem prudent to use a battery of techniques to confirm or refute the presence of transferrin receptors rather than using only monoclonal antibodies to the receptor.

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Dr Gatter replies as follows:

Dr Wells and colleagues report the interesting observation that a different monoclonal anti-transferrin receptor antibody to the four that we used in our study does not give staining of the basal layer of the epidermis in their laboratory. We too pointed out that there were differences between our antibodies, particularly between BK19-9 and the other three antibodies. The latter half of our discussion was concerned with the possible explanation for this and we raised the possibility that these discrepancies might reflect the fact that the transferrin receptor is not a single molecular entity but a family of molecules which are antigenically similar but not identical. Further studies, using a variety of different techniques, on the human transferrin receptor will be of great interest.

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Selective damage to type 2B muscle fibres in ethanol-fed rats

The recent report by Slavin *et al*¹ describes selective atrophy of the alkaline myofibrillar ATPase type 2B muscle fibres in chronic alcoholics. Slavin *et al* suggest that

defective muscle anaerobic glycolysis may underlie the selective damage to type 2B fibres that are more heavily dependent upon glycolysis for energy production. The prototype disorder of defective skeletal muscle anaerobic metabolism is myophosphorylase deficiency (glycogenosis type V, McArdle's disease), although phosphofructokinase deficiency (glycogenosis type VII, Tarui's disease), phosphoglycerate mutase deficiency (DiMauro's disease), muscle lactate dehydrogenase deficiency (Kanno's disease) and muscle phosphoglycerate kinase deficiency (Dalakas' disease) have similar symptomatology.^{2,3} These disorders are all characterised by painful, exercise-induced, electrically-silent muscle contraction followed by post-exercise rhabdomyolysis.^{2,4} The usual diagnostic clinical test, the forearm ischaemic exercise test, is based upon the finding in these disorders of deficient lactate production during ischaemic exercise.⁵ Radionuclide scanning with calcium tracers demonstrates marked uptake in contracted muscle.^{6,7} Similar symptomatology including exercise-induced, painful muscle cramping, episodes of rhabdomyolysis, excessive muscle uptake of radionuclide calcium tracers, and defective lactate production during ischaemic exercise has been reported in chronic alcoholics.⁸⁻¹¹ In our histological studies of patients with myophosphorylase deficiency or phosphofructokinase deficiency, we found selective damage to the type 2B muscle fibres.^{4,6}

We have developed an animal (rat) model for disorders with defective skeletal muscle glycolytic/glycogenolytic metabolism.¹²⁻¹⁴ This model utilizes iodoacetate selectively to inhibit the second stage glycolytic enzyme D-glyceraldehyde-3-phosphate dehydrogenase. The animals develop muscle symptomatology completely analogous to that of the human patients, including the histological evidence of selective type 2B muscle fibre injury. We have also found a sexually dimorphic response in this model with male rats and ovariectomised female rats developing markedly more severe symptomatology than intact female rats.¹⁵

We have performed preliminary studies of the effect of ethanol in the animal model. For six to ten weeks a complete liquid diet (Ensure) to which was added 9.5% ethanol by volume was administered as the only nutrient to a group of Wistar-Furth rats. Although these rats show no evidence of alcohol dependence, they do have persistent behavioural deficits.¹⁶ After administration of less than half the usual